

Grant# HD44404

Center Director: Jashvant D. Unadkat, Ph.D.

Center Overview

Pregnant women and their fetuses are therapeutic "orphans." As recently stated by Dr. Kennedy of the FDA, now it's time to provide the best information we can for women who take drugs while they're pregnant." This SCOR application seeks to fulfill this national goal. Drugs are administered to pregnant women, and therefore their fetuses, without the necessary clinical data about the pharmacokinetics, dose, safety, or efficacy of the drugs in these vulnerable populations. To determine the correct dose of a drug to administer to the pregnant woman, it is important to know if the pharmacokinetics of the drug is different in pregnant women when compared with men or nonpregnant women. In this SCOR application we will focus on one of two major routes of drug disposition, namely drug transport. We will test the following overarching and unifying hypothesis: Expression and activity of influx and efflux transporters is upregulated during pregnancy to respectively meet the increased need of nutrients by the mother and her fetus and to minimize toxicity from xenobiotics. The transporters we will study are those likely to be quantitatively most important in drug transport in both the Intestine and the placenta, namely P-glycoprotein (P-gp; Project 1), the Breast Cancer Resistance Protein (BCRP; Project 2), and the Organic Cation Transporter 3 (OCT3; Project 3). In the placenta, P-glycoprotein and BCRP participate in the efflux of drugs, thus protecting the fetus from deleterious effects of drugs, while OCT3 functions to allow the entry of endogenous compounds (and drugs) into the fetal circulation. In contrast, all three transporters play a synergistic role in the elimination of drugs in the liver and intestine. Moreover, all three transporters have overlapping but distinct substrate specificity, transporting a wide variety of drugs of diverse therapeutic categories. Studies outlined in the three projects will elucidate the interplay among these three transporters in modulating maternal AND fetal exposure to drugs. Therefore, the three projects will pursue a single goal of elucidating mechanisms by which drug, transporters alter maternal and fetal drug exposure during pregnancy. Results obtained from these studies will have wide-ranging consequences for drug use during pregnancy. First, based on the data we obtain, we will be able to predict which drugs routinely administered to pregnant women are likely to have their disposition affected by pregnancy. Second, our data should allow predictions on the magnitude of change likely to be observed. In a future application, we will test these predictions in the clinic. Once tested, we will be able to predict the magnitude of change in dose that should be instituted for a wide range of drugs whose disposition is significantly modulated by these transporters.

Principal Investigator: Jashvant D. Unadkat, Ph.D.

Project 1: Hepatic/Intestinal P-glycoprotein and CYP3A in Pregnancy

Pregnant women and their fetuses are therapeutic "orphans." Drugs are administered to pregnant women, and therefore their fetuses, without the necessary clinical data about the pharmacokinetics, dose, safety, or efficacy of the drugs in these vulnerable populations. To determine the correct dose of a drug to administer to the pregnant woman, it is important to know if the pharmacokinetics of the drug are different in pregnant women when compared with men or non-pregnant women. Many drugs administered to pregnant women are substrates of P-glycoprotein (P-gp) or cytochrome P450 3A enzymes (CYP3A4/5) or both, such as antivirals (e.g., anti-HIV protease inhibitors, antibiotics (e.g., clarithromycin), antihistamines (e.g., fexofenadine), and anti-epileptics (e.g., carbamazepine). P-gp and CYP3A4/5 enzymes are strategically located in the intestine, liver and kidneys--organs important for absorption, metabolism, and excretion of drugs. We have recently obtained evidence from perinatal Phase I clinical trial on indinavir, an anti-HIV protease inhibitor, that the oral clearance of this drug is increased approximately 3-fold during pregnancy. Since the disposition of indinavir is determined by P-gp and CYP3A4/5 enzymes in the intestine and liver, we hypothesized that P-gp and CYP3A4/5 expression and activity in these tissues are enhanced during pregnancy. The Specific Aims listed below are designed to test this hypothesis.

Hypothesis:

Hepatic and Intestinal P-glycoprotein and CYP3A4/5 expression and activity is enhanced during pregnancy

Specific Aims:

1. To determine, both antenatal and postpartum, in vivo intestinal and hepatic P-glycoprotein and CYP3A4/5 activities in pregnant women by oral administration of selective substrates of P-gp (digoxin) and CYP3A4/5 (midazolam).
2. To determine, in vivo (or ex vivo), both antenatal and postpartum, intestinal and hepatic P-glycoprotein and CYP3A4/5 activities (or expression) following oral and IV administration of protease inhibitors to a representative animal model, the pregnant M. nemestrina.
3. To determine if activity and expression of P-gp in lymphocytes is elevated during pregnancy in women and M. nemestrina.

Principal Investigator: Qingcheng Mao

Project 2: BCRP in Pregnancy: Activity, Expression and Regulation

While the importance of P-glycoprotein (P-gp) in determining drug disposition has been well recognized, the role of Breast Cancer Resistance Protein (BCRP) in this regard has just begun to be realized. Both P-gp and BCRP are expressed in the apical membranes of small intestinal epithelium, the liver canalicular membranes, and the placental syncytiotrophoblasts. Thus, it is not surprising that BCRP, like P-gp, affects the bioavailability and fetal distribution of drugs. Pregnant women are routinely administered various drugs such as antivirals, anti-epileptics, antibiotics, anti-hypertensives, and anti-histamines. Many of these drugs are substrates or modulators of P-gp. As BCRP and P-gp have considerable degree of substrate overlap, many of these drugs are also likely to be substrates of BCRP. In order to assess if BCRP plays an important role in determining the safety and bioavailability of drugs given during pregnancy, it is critical that we first determine which of the drugs routinely administered to pregnant women are substrates of BCRP. BCRP has the highest expression levels in the placenta where it functions, like P-gp, to protect the fetus from Xenobiotics. Preliminary data from our laboratories indicate that expression of both BCRP and P-gp in the placenta is gestational-age dependent, suggesting regulation by pregnancy-specific hormones. The Hypothesis and Specific Aims outlined below are designed to address these clinically relevant questions.

Hypothesis: BCRP and placental P-gp activity and expression is up-regulated during pregnancy and alters the absorption, distribution (including across the placenta), and elimination of drugs BCRP and P-gp substrates) routinely administered to pregnant women.

To test the above hypothesis we will determine:

1. If drugs routinely administered to pregnant women (e.g., anti-HIV protease inhibitors, anti-epileptic drugs, antibiotics, anti-hypertensives and antihistamines) are high-affinity substrates of BCRP.
2. If in vitro and in vivo expression of BCRP and P-gp is regulated by pregnancy-specific hormones.
3. The molecular mechanism by which BCRP and P-gp expression is regulated by pregnancy-specific hormones.
4. If the in vivo absorption and fetal distribution of a high-affinity BCRP substrates (routinely administered to pregnant women) is affected by pregnancy in P-gp-deficient mice.

Principal Investigator: Vadivel Ganapathy, Ph.D.

Project 3: OCT3 in Drug Pharmacokinetics During Pregnancy

The aim of this project is to understand the role of OCT3 (organic cation transporter 3), an important drug transporter in the placenta, in the pharmacokinetics and fetal exposure of commonly used therapeutic drugs during pregnancy in normal women and in women infected with the AIDS virus HIV-1. OCT3 belongs to a larger family of drug transporters that handle a variety of drugs and other xenobiotics. Among the members of this gene family, OCT3 is the most relevant to the handling of drugs in women during pregnancy because it is the only OCT subtype that is expressed in the placenta. There have been several interesting and intriguing recent findings regarding the function and expression of OCT3 during pregnancy and its regulation by the HIV-1 protein Tat. These findings form the basis for the following hypotheses: a) OCT3 is expressed on the maternal side of the placental syncytiotrophoblast where it enhances fetal exposure to a wide range of therapeutic drugs; b) The placental expression of OCT3 increases with gestational age and consequently OCT3-dependent pharmacokinetics and fetal exposure of therapeutic drugs and other xenobiotics vary significantly at different stages of pregnancy; c) The expression of OCT3 in non-placental tissues is influenced by pregnancy and type 1 sigma receptor and hence the pharmacokinetics of OCT3-specific drugs is significantly altered in women during pregnancy; and d) The expression of OCT3 in placenta and other tissues is down-regulated by HIV-1 Tat and therefore the role of OCT3 in pharmacokinetics and fetal exposure of therapeutic drugs is altered markedly in women infected with HIV. Studies are proposed in this project to test each of these hypotheses. Studies related to the expression and polarized distribution of OCT3 in the placenta at different gestational ages will be carried out using human placentas. Differential polarization of OCT3 in the placenta versus the kidney and intestine and the influence of progesterone and the type 1 sigma receptor in the regulation of OCT3 expression and function will be investigated using appropriate human cell lines. Substrate specificity studies with emphasis on the therapeutic drugs that are commonly used in women during pregnancy will be done with the cloned human OCT3. The role of OCT3 in the pharmacokinetics and fetal exposure of therapeutic drugs and its modulation of HIV-1 Tat during pregnancy will be assessed using wildtype, OCT3^{-/-} knockout, and Tat-transgenic mice. These studies will generate clinically and therapeutically relevant new information on the role of OCT3 in the handling and fetal exposure of drugs and other xenobiotics in normal women and in women with AIDS.